Note on Eggs and Embryos of the South African Myxinoid, Bdellostoma (Heptatretus) hexatrema, Müll.

By

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With Plates 10–12.

The problem of the exact relationship of the Cyclostomes to Amphioxus on the one hand and the Gnathostomes on the other has been rendered more difficult of solution on account of the lack of knowledge of the early stages of the Myxinoids. While the Lampreys have been very thoroughly investigated in this respect, there remains some considerable obscurity with regard to the details of the development of the Hag-fishes. Of the two chief genera, Myxine and Bdellostoma, embryological material has only been procured in the case of the latter, and that so inadequately that a detailed comparison with other groups of animals is not yet possible. Sufficient, however, is known to show that the development differs essentially from that of the Lampreys, thus rendering it all the more necessary that a fuller knowledge of it should be obtained.

Various species of Bdellostoma are found at widely separate localities—California, Alaska, Chile, South Africa, New Zealand, and Japan—and, though they occur in abundance at such places, their eggs and embryos are little known. In the case of the Californian B. stouti our knowledge of early stages has been considerably advanced by Dean (2), Price (6), and others. Eggs and some embryological material are recorded from Japan by Dean (**3**). The eggs of a species of Bdellostoma are also recorded from Chile (**5**), (**7**). Otherwise, so far as I am aware, no other material of this kind has been procured. The discovery, therefore, of some naturally deposited eggs with embryos of the South African species is of interest. Repeated attempts to find these have hitherto been in vain. The probable appearance of such eggs has been described to fishermen and others likely to have come across them, but none knew of such objects having been found in the sea. Recently, however, Mr. Cripps, the Cape Province Fishery Officer, found and recognised the eggs from the description given, and placed them in methylated spirits, the only preservative available at the time.

The eggs, five in number, were found on August 23rd, 1916, in a small bay ("Fiddle Bay") on the west coast of South Africa, in which were rocks interspersed with mud and sand. They were found near low water mark, partly embedded in mud, two of them only being visible on the surface. It proved on examination that these two alone had any contents; those embedded in the mud were empty, their contents having been removed in all probability by some predaceous animal, for in each of the empty shells there was a small, irregular aperture, with edges torn in such a way as might have been done by the radula of a mollusc. These openings are seen in two eggs in Pl. 10, fig. 1, from a photograph of the eggs as received. The eggs were attached to each other by their anchoring filaments, the attachment having been rendered more secure by the presence of the cast-off byssi of some bivalve. The threads of these were in some cases attached to the surface of the egg, and in others to small pebbles. These byssi, two or three of which were found at the end of each egg, were probably those of Mytilus crenatus, Lam., a small specimen of which, 18 mm. in length, was attached at the point of junction of three egg-cases. All these circumstances seem to point to the fact that the group of eggs had been detached from the place in which they were naturally deposited.

Two of the eggs (Pl. 10, fig. 1, *a* and *e*), which were not embedded in the mud, contained embryos in an advanced stage. These were at each end of the chain. One (*a*) has been cut open, and the contained embryo placed alongside of it, as shown in the photograph.

Dimensions of Egg.

The five eggs are very uniform in size, being about 30 mm. in length and 12 mm. in greatest breadth. This is somewhat longer than those of Bdellostoma stouti, which are stated by Dean to range from 14·3 to 29 mm., and decidedly larger than the eggs of the Bdellostoma from Chile, which were 25 mm. at most. Müller records an ovarian egg of a Bdellostoma from the Cape of Good Hope 31 mm. in length, and I have confirmed this by examination of well-advanced ovaries, in which some eggs were even 33 mm. in length, but much narrower than the naturally deposited egg. The extra three millimetres in the ovarian egg can hardly be looked on as casting doubts on the identity of the present eggs, and this is conclusively proved by the characteristics of the embryo.

The breadth of the egg (12 mm.) is decidedly greater than that of B. stouti, which varies from 6·8 to 10·5 mm.

Shape of Egg.

The egg-shell is slightly bent on its long axis. This is most clearly shown by the fact that if the planes of the two polar rings, which are approximately at right angles to the long axis of the egg, were produced, they would meet at a distance from the egg of about two or three times its length. This asymmetry of the egg appears also very distinctly in well-developed ovarian eggs, the concave or straight side being sometimes next the wall of the mesovarium, but more frequently on the opposite side. Dean, on the other hand, has not found in B. stouti a notably asymmetrical ovarian egg, nor has he seen this asymmetry in newly deposited eggs, and he is inclined to believe that the asymmetry may be due to the difference in physical characters of the two sides of the outer egg-membranes, and acquired after deposition.

General Structure of the Shell.

The shell shows a number of small, superficial markings, usually circular or polygonal, and of varying sizes. They may, however, vary considerably in shape, some being elongate, and occasionally elongate and bent on themselves. That these markings may assume an elongate form is of special interest, as will be seen later on. The markings are brought about by the hard and dark brown outer surface dipping down into the substance of the shell so as to appear superficially as dark lines, and in sections as a number of short columns. These are continued inwards as colourless columns, and constitute the zona striata described by Cunningham (1) and Mark (4) in Myxine and by Dean (2) in Bdellostoma. Dean uses the name "villi" and "filaments," but it will be more explicit to use here the term "columns" for these structures, and, for their terminal pigmented ends, the term "heads of columns."

Below this columnar layer there is, as in B. stouti, a layer of stratified material, consisting of from seven to nine broad bands or strata with finer lamellations, and below this stratified layer is a thin but tough homogeneous layer, in which, at least in the greater part of the egg-shell, no differentiation can be seen.

The following regions of the shell may therefore be recognised: (1) The heads of the columns; (2) the columnar layer; (3) the stratified layer; (4) the homogeneous layer. A useful distinction also is that the first and the last, to a lesser extent, are of a yellowish or dark brown colour. The third, or stratified layer, is further distinguished from the others in chemical composition, as it can readily be dissolved when boiled in caustic potash. These distinctions, it will be seen, are necessary, as the different layers occur in very different proportions in the egg-shell of the Cape Bdellostoma.

Papillæ or Projections on the Shell.

Scattered over the surface of the shell are a number of minute, projecting points or papillæ, which can be seen in the dry condition with the naked eye. About fifty were counted, with the aid of a lens, across the shell in one case. They occur all over the egg, close to and on either side of the opercular ring. That these small projections are made up of a number of columns, and not of one enlarged column, is readily seen from the fact that the polygonal markings can be traced from their basis to their tips (Pl. 11, fig. 3). The tips by transmitted light appear as clear points under the microscope, and the dark lines forming the polygonal markings end abruptly round the periphery of the clear spot or are continued on it as faint lines or disconnected dots. A section through the projection (Pl. 11, fig. 4) shows that its formation is due to the increase in thickness of the stratified layer. The columns towards the centre of the projection are somewhat crowded together, and, at their distal end, are mostly devoid of the dark pigment characteristic of the heads of the columns elsewhere, thus giving rise to the clear spot seen by transmitted light in a surface view.

The Opercular Rings.

The eggs differ from those of B. stouti and those recorded from Chile by Putnam (**7**) in having an opercular ring at each end of the egg; in this respect, however, agreeing with the eggs recorded from Chile by Plate (**5**) and from Japan by Dean (**3**). The opercular rings are not equally developed. One is well marked in all the eggs, and is situated about 6 mm. from the end of the egg. The other is well marked in three of the five specimens, but in one (Pl. 10, fig. 1, *e*) only a slight trace of it can be found. In specimen *a* there are two well-marked rings, one 4·5 mm. from the terminal filaments, the other only 1 mm. from the filaments; in specimen *b* the rings are also well marked, the one being 4 mm. from the terminal filaments, the other 1 mm.; in *c* the distance is again 4·5 mm., but there is only a very indistinct mark of a second ring ·5 mm. from the filaments; in *d* there are two rings, one well marked, and 4·5 mm. from the filaments, the other imperfect, being evident on one side only, where it is 1 mm. from the filaments; in *e* there is again a well-marked ring, about the same distance from the filaments, but there is no definite second ring, though there is a faint indefinite mark, about 1 mm. from the base of the filaments.

The rings appear in a superficial view as dark brown lines or bands forming a slight ridge round the egg-shell. The polygonal markings occur on either side, but, as they approach the ridge, they become somewhat broken up into lines and dots, and on the ridge itself they cannot be seen, just as in the case of the tips of the shell-projections.

The diameter of the large opercular ring is very constant, being about 10 mm., while that of the smaller varies from 5·5 mm. to 7 mm. These measurements are, of course, no indication of the shape of the egg, for the smaller ring is nearer the end.

The structure of the opercular rings is of special interest. In B. stouti they do not differ essentially in structure from the rest of the egg-shell, for, as Dean (**2**) states, in the process of rupture at the ring a slight fissure first appears between the bulbous tips of the outer or columnar layer; this extends downwards between the columns, and, about the time of hatching, passes into the middle stratified layer, and the process of warping, thus brought about, doubtless causes rupture of the inner non-striate membrane. In the present material, however, the rings are of a much more definite nature.

Their structure may be best made out by first examining sections of varying thickness without special treatment, and then observing the changes brought about by treatment with caustic potash. Results may be obtained more quickly by using hot or boiling solution, but it was found more instructive to observe the changes brought about by prolonged reaction. In thick unaltered sections in the long axis of the egg the ring appeared as a broad band or pillar through the substance of the shell, and of the yellowish colour characteristic of the outer part of the shell. This part, the heads of the columns (Pl. 11, fig. 5, *h. col.*) is somewhat different here, as was also observed in a surface-view, in which dark lines were broken up or fragmentary. The columnar layer has changed its character, being much reduced or absent, as no distinct columnar structure was observed here. The stratified layer has, however, greatly increased in thickness, and extends upwards almost to the heads of the columns. At the point where the strata meet the pillar, they are bent outwards, and in thin sections they appeared to end here; in thicker sections, however, they were seen to be continued across the pillar (Pl. 11, fig. 5). If now the section be treated with caustic potash, the stratified layer is entirely dissolved away, and the structure of the pillar becomes much more evident. It consists of a clear structureless groundwork similar to the substance of the homogeneous layer, with which it is continuous. It becomes narrower towards the outer surface, where it appears to merge into the layer of the heads of the columns, though it cannot be distinctly followed here. Running up the centre of this pillar is a very distinct dark brown line, which meets the layer of heads of the columns at its distal extremity, and, at its proximal extremity, passes into the homogeneous layer. On each side of this line there are others running parallel to it, and of the same colour, but much thinner. They do not reach the outer surface, but, towards the inner surface, they become more numerous, and finally spread out at the proximal end, and merge into the homogeneous layer.

It appears from these observations that the ring-structure is brought about by the great development of the homogeneous layer, as a consequence of which the stratified layer, though carried somewhat outwards, has not diminished, but rather increased in breadth, leading to a reduction or disappearance of the columns of the columnar layer, and a modification of the heads of the columns. The brown line, which runs across the shell from inner to outer surface, and the thinner lines on each side of it, suggest the structure of the columnar layer, but they are more intimately connected with the homogeneous inner layer. Functionally it is doubtless connected with the rupture of the shell and the throwing-off of the terminal capsule at the time of hatching.

Structure of Polar End of Shell.

The polar ends of the shell consist of the same layers as the body of the shell, but in such different proportions and arrangement that they require special notice.

From the ring towards the apex of the shell the homogeneous layer is at first in the form of a thin almost colourless membrane, but increases somewhat abruptly in thickness (Pl. 11, fig. 6) and assumes a yellow tinge, till near the apex it is the most conspicuous part of the shell-structure (Pl. 11, fig. 7). Stout offshoots are given off from it to the polar filaments, and, at the micropylar region, it constitutes practically the whole thickness of the shell. It still retains its homogeneous structure, which, however, becomes slightly granular in parts.

The stratified layer does not increase much in thickness, but it comes to lie near the outer aspect of the shell, and it is interrupted by the above-mentioned offshoots of the homogeneous layer, so that in section it appears more or less in patches. It may be seen to run up into the bases of the filaments (Pl. 11, fig. 7).

The columnar layer has become very much reduced, and at most places the columns have disappeared, but their hard-coloured heads are well-developed, though somewhat modified at places, as noted below.

The Micropyle and Micropylar Funnel.

The micropyle is not a simple canal, but is composed of definite and well-marked parts (Pl. 11, fig. 7, *mp.*). It lies in the homogeneous layer. The distal end, by which it opens to the exterior at the base of the wide micropylar funnel (*m.f.*), consists of a well-defined straight tube narrower than the rest of the tube. It then expands into a more or less spherical dilatation, and is continued to the inner surface as a tube, somewhat wider than the first portion. Where it meets the inner surface of the shell, it expands into a funnel-shaped opening.

The micropylar funnel (Pl. 11, fig. 7, *m.f.*) is very similar to that in B. stouti, being cup-shaped ; it cannot be described strictly as a funnel, as the bottom is somewhat flattened, as in this species and also in Myxine (**1**). It is wider than in B. stouti, being about ·4 mm. in greatest diameter as against ·25 mm. in this species. Its depth cannot be accurately determined, as its sides pass gradually into the bases of the surrounding filaments. The base and part of the sides are made up of the homogeneous layer, but, on its sides, may be seen in surface-view the polygonal markings, which occur in a more or less modified condition over the whole surface of the egg. They probably also occur on the base, but a surface-view of this region was not obtained. The markings are very superficial, and do not apparently extend inwards to the underlying layer. The funnel was filled with a structureless extraneous substance, the nature of which was not determined. This substance was also found forming a hard white encrusting layer on the shell between the bases of the filaments (Pl. 10, fig, 2).

The Anchor-Filaments.

The anchor-filaments of the Cape Bdellostoma are characterised by their shortness, the longest being 3·5 mm. They are, however, more numerous than in other species, there being about a hundred at each end. They are arranged more or less concentrically at the animal pole of the egg (Pl. 11, fig. 8). In the specimen drawn there were 117 of these filaments.

The outer filaments are shorter, and some are mere projections, apparently homologous with the projections on the body of the shell, and have no anchors. In most cases, however, those without anchors have had them broken off.

There appears to be some considerable difference of opinion as to the nature of the anchor-filaments. A comparison of the egg-case and its filaments with those of Elasmobranchs, suggested by Thompson and considered unfavourably by Putnam and Cunningham, has little to justify it. Dean puts forward a very definite suggestion, which seems to be supported by the material at his disposal. It is that "the anchor filaments are homologous with the villi of the outermost (definitive) shell layer, zona striata. The anchor represents the highly-specialised bulb of the villus, its stalk the filament of the latter." In other words, the anchor-filament is a greatly enlarged and specialised column of the columnar layer, and the anchor is its head. The facts in favour of this view seem to be that in B. stouti no trace of the villi are found in or on the shell immediately surrounding the filaments, and that the filament with its anchor is constituted of the same elements as the villus or column of the columnar layer. Thus the filament was found to be homogeneous in its structure, with no trace of the stratified layer. There were, indeed, some fine striations found on the surface, but as these were merely superficial, and visible only under a high power of the microscope, no particular importance was attached to them. The surface of the anchors showed no pits, dots, nor irregularities, being smooth like the surface of the bulbous tip of the villus.

The egg-cases of the Cape Bdellostoma differ from those of the Californian species in these particulars, and this can be seen without minute examination by sectioning. Thus the polygonal markings, indicating the heads of the columns, can be clearly seen extending up over the surface of the egg-case towards the polar end and between the bases of the outer filaments. They extend on these bases, and, if such a filament and its base be cut out and flattened under a coverglass (preferably after boiling in caustic potash) so as to obtain a clear surface-view, it is seen that the markings extend up over the base to the stem of the filament (Pl. 11, fig. 9). From the base upwards they, however, gradually change from a polygonal to a more elongate shape, till, on the stalk itself, they appear as elongate striations. These are not specially fine, and can readily be seen with a Zeiss A objective. Towards the anchor they divide into groups of two or three according to the number of anchor-lips or projections; they spread out over the under side of these and end abruptly at their margins (Pl. 11, fig. 10). Between the bases of the more closely set inner filaments the markings were not polygonal, but somewhat elongate, as on the filaments themselves. The striations on the filaments are therefore apparently modified polygonal markings, and are the drawn-out heads of the columns.

If, now, the distal surface of the anchor be examined, no striations are seen, but a number of dark spots are very clearly visible, extending, not only over the central part, but on to the upper surface of the projecting parts or flukes of the anchor, where, however, they become fainter (Pl. 11, fig. 10). They suggest similar markings seen on the projections and the rings of the egg.

All this can be seen without more minute examination, but is made much clearer in sections. Thus a longitudinal section through the filaments shows that not only the stratified layer, but also the homogeneous layer, passes up into the bases of the filaments (Pl. 11, fig. 7). These branches of the homogeneous layer passing into the filaments are very conspicuous in such sections owing to their brownish colour, and were noted by Dean, who, however, describes them as the continuations of the bases of the filaments into the substance of the shell. Transverse sections of the filament itself (Pl. 12, fig. 11) indicate clearly that these elements are continued up into its main stem. The columnar layer is here well developed, though the caps of the columns are not so well marked. Internal to this is the stratified layer, and, occupying the centre, the homogeneous layer. The elements were seen only after prolonged treatment with caustic potash or by slight heating. In untreated sections the filament appears to be solid and structureless, as Dean describes them in *B. stouti*, and, if actually boiled in the solution, unless the section is very thick, nothing is left except the outer striated layer representing the heads of the columns, as Dean describes and figures (his fig. 13).

Sections of the anchor (Pl. 12, fig. 12) show that here, also, the stratified layer is present, and constitutes the main body of this part, the outer surface being made up of the harder heads of the columns. The hook-like projections or flukes of the anchor are difficult to section, but they are apparently made up entirely of the heads of columns, which would account for their hard, tough nature. Their upper surface shows a number of dots, similar to those of the upper surface of the anchor generally, and their lower surface shows the striations characteristic of the surface of the filament.

The reasons for believing that the filament represents a modified villus or column of the zona striata, and the anchor its terminal bulbous end or head, seem to be convincing enough in the case of the Californian *Bdellostoma*, but, as is apparent from the above description, these reasons do not exist in the case of the Cape species, in which both the filament and its anchor consist of the columnar and stratified layer, along with the homogeneous layer (in the filament, but apparently not in the anchor). It may be suggested that the condition in the Californian species is a specialisation and modification of the more primitive condition found in the Cape species, and that the homogeneity of the filament and anchor are secondarily acquired. The fine striations in the filament noted by Dean may be significant in this respect.

Respiratory Apertures of Egg-Case.

Though the embryo of *Bdellostoma* is abundantly supplied with yolk, and presumably has, like the yolk-laden embryos of Elasmobranchs, a comparatively long period of development, there are are no special embryonic respiratory organs, as in this group. More remarkable is the fact that there is apparently no means of introducing an adequate current of water for respiratory purposes, as in the Elasmobranchs. The absence of special embryonic respiratory organs may be compensated for by the highly vascular nature of the surface of embryo and yolk, or the functional activity of the gill-pouches at an early stage; but only somewhat uncertain indications of canals through the egg-membranes have been detected. It can hardly be doubted that a free supply of water must in some way pass through the tough egg-case, yet Dean could find no clear proof that canals passing through the shell existed. In the case of the Cape Bdellostoma no clearer indications of canals passing through the egg-case were seen, but there were numerous longitudinal fissures, some appearing as mere cracks in the surface, others passing completely through the egg-shell (Pl. 10, fig. 2). They became fewer near the opercular rings, and were not so marked on the polar side of the rings.

The mechanism whereby water can be drawn in through the fissures may well be the movement of the much-flattened body, though in the advanced embryo there is little space between the egg-case and the yolk in which this can take place.

External Characters of Embryo.

Of the two embryos one is slightly more developed than the other; both extend completely round the egg in the direction of its long axis; in one the tail overlaps the head-region by about one-third of the length of the egg (Pl. 12, fig. 13), while, in the less advanced one, this overlapping was only about one-fifth. The egg and embryo form a mass, which, like the egg-capsule, is somewhat curved, the more curved side representing the back of the embryo, which is still attached to the yolk. On the ventral side the embryo is quite free from the yolk, the point of separation, both in the case of the free head-region and the tail, being the extremities of the yolk. These free portions of the embryo are, however, closely applied to the yolk, and lie in a shallow, wide furrow on itsventral aspect. Both portions are much compressed, and are somewhat twisted on themselves at the points where they become free from the yolk. This twisting is more marked in the tail-region, which comes to lie flat on its left side against the yolk. The compression thus is from side to side, so that the mucous sacs of the right side are visible on a surface-view. The anterior and longer free end of the embryo does not, however, undergo so much torsion, and this region is flattened, not laterally, like the tail-region, but dorso-ventrally, so that the mucous sacs are applied to the yolk, and are not visible from a superficial view. That there is a slight twisting, however, is shown by the fact that the mucous sacs of the right side are much closer to the margin of the body than those of the left.

These sacs are very prominent at this stage, and appear as little hillocks. They are specially well marked on the yolk, where about forty were counted. Here they lie about midway between the dorsal and ventral margin of the egg, being somewhat nearer the dorsal, so that they are visible in a dorsal, but not in a ventral view of the egg. They are thus further removed from the main axis of the embryo than in Bdellostoma stouti.

The length of the anterior free end from snout to the last gill-opening is 26 mm.; the length of the middle portion, which is attached to the yolk, is 21 mm.; the length of the posterior free end, from anus to end of tail, is 15 mm.; the total length of the embryo is, therefore, 62 mm.

Structure of Embryo.

As only two embryos were procured, it did not seem advisable to sacrifice more than one for detailed examination and sectioning. Owing to the advanced stage of these embryos, it was not to be expected that they could throw much further light on the development of the Myxinoids, and, so far as an examination has shown, the embryo, unlike the shell, does not exhibit any primitive or specially instructive features. For an adequate examination a complete series is desirable, and this will doubtless be procured at some future time. One or two points, however, may be noted in connection with certain features of this stage.

The anterior free end of the embryo, when stained and viewed from the ventral aspect, shows that the various organs are well developed. Three pairs of tentacles and the tentacular skeleton are practically as in the adult; the cartilages of the naso-pituitary canal and of the olfactory capsule are well developed. The notochord is seen in transverse section to be much compressed from above downwards, and is apparently at this stage a very flexible organ, adapting itself readily, not only to the flattening, but also to the twisting of the body at the tail-region.

The condition of the excretory system is, however, of more interest at this stage. Its main features can readily be made out in a stained preparation. The pronephros measured about ·58 mm. in length, and occupied about one and a half segments of the body, judging by the position of the spinal ganglia. The mesonephros, consisting of a long segmental duct and twenty-two tubules with their glomeruli, was clearly made out. It is believed that the Myxinoids differ from all other Craniates, in that they alone have the mesonephric tubules strictly segmental. In this embryo, however, the anterior tubules were obviously much nearer each other than the more posterior, and were apparently not strictly segmental. This was confirmed by a series of sections, which showed that, between the posterior end of the pronephros and the first following spinal ganglion (right side), there were two tubules opening into the segmental duct; between this and the second spinal ganglia two tubules open into the segmental duct, and two between the second and third ganglia, there being thus six mesonephric tubules in three segments of the body behind the pronephros. Posterior to the sixth tubule of the mesonephros, there was one tubule to each segment of the body to the end of the mesophrones.

In front of the first tubule of the mesonephros is a tubule of special interest (Pl. 12, fig. 14, tu_1.). It appears in the stained preparation as a somewhat elongate straight tube, passing from the segmental duct into the pronephros, where it ends in a glomerulus. At the point where this tubule touches the segmental duct, the latter is interrupted (Pl. 12, fig. 14, *s.d.*), though immediately in front of and behind this point it has a complete lumen. There is, however, no distinguishable communication between the tubule and duct as in the tubules of the mesonephros. In front of this tubule and now completely in the pronephros is another (Pl. 12, fig. 15, tu_2.) of a similar nature, connected proximally with another glomerulus, and distally ending near the segmental duct, which is, however, now solid (Pl. 12, fig. 15, *s.d.*). The third tubule of the pronephros is also attached proximally to a glomerulus, and distally seems to end blindly. At the bases of the other tubules is a mass of vascular tissue, which might be the result of fusion of a number of glomeruli, while, at the distal end, they open by large nephrostomes into the body-cavity, the segmental duct having disappeared.

No undoubted trace of the further continuation of the segmental duct into the pronephros, after its disappearance at the distal end of the second last tubule, was discovered. Though the tubules sometimes expanded into wide cavities (Pl. 12, fig. 15 *tu.*), and were connected with each other, there appeared no sufficient reason to regard these as traces of the segmental duct (cf. Price (**6**)).

Summary.

(1) Five naturally deposited eggs of the Cape Bdellostoma have been found, two containing well-advanced embryos.

(2) The eggs are larger than those of other species, the anchor-filaments are shorter, and there are two polar rings.

(3) The general structure of the shell is similar to that described for other species, and there are numerous small projections on its surface, as in some species. These consist

of the columns of the columnar layer modified at the apex of the projection.

(4) There are numerous small fissures in the shell, probably respiratory apertures.

(5) The polar rings have a definite structure, differing from that of the rest of the shell, in that the inner layer becomes greatly enlarged, and the outer layer much reduced.

(6) The anchor-filaments are not homogeneous in their structure, but consist of all the layers of the shell, the chief modification being that the heads of the columns of the columnar layer becomes drawn out so as to appear as striations.

(7) The anchors consist of the modified columnar layer and the stratified layer. On their outer surface the heads of the columns of the columnar layer appear as disconnected dark dots, while their lower surface consists of the same elements as the surface of the filament.

(8) In the embryo the segmental duct occurs at the distal end of the last tubule of the pronephros, but, though having a lumen, does not open into it. It is found also at the distal end of the second last tubule, where, however, it becomes solid, and disappears. It was not found extending further into the pronephros.

(9) The tubules of the mesonephros are not strictly segmentally arranged, in that there are six tubules in three segments of the body behind the pronephros, though there is one tubule for each succeeding segment, as far as the mesonephros extends.

I am greatly indebted to Prof. Bashford Dean for his generous assistance in literature on the early stages of Bdellostoma and allied subjects. As a pioneer in this work, he is deeply interested in the finding of the eggs of the Cape Bdellostoma. Prof. Price has also kindly sent me reprints of his important papers on the development of the excretory organs.

I have also to express my obligations to Mr. P. MacManus, who has redrawn for me figs. 2 and 10.

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EXPLANATION OF PLATES 10–12.

Illustrating Dr. J. D. F. Gilchrist's "Note on Eggs and Embryos of the South African Myxinoid, Bdellostoma (Heptatretus) hexatrema, Müll.

PLATE 10.

Fig. 1.—Photograph of eggs of Bdellostoma as received. *a–e*. The eggs. a^1. Embryo removed from *a*.

Fig. 2.—Egg showing papillæ, polar rings, anchor-filaments and fissures.

PLATE 11.

Fig. 3.—Polygonal markings on surface of egg up to clear surface of papilla.

Fig. 4.—Vertical section through papilla. *col. l.* Columnar layer. *hom. l.* Homogeneous layer. *pap.* Papilla. *str. l.* Stratified layer.

Fig. 5.—Section across polar ring. *col. l.* Columnar layer. *h. col.* Heads of columns. *hom. l.* Homogeneous layer. *str. l.* Stratified layer.

Fig. 6.—Longitudinal section through shell of egg near outer filaments, showing change in layers. *f.* 1. Filament cut at side. *f.* 2. Filament cut near middle and broken.

Fig. 7.—Longitudinal section through apex of shell showing micropyle. *f.* Filament. *h. l.* Homogeneous layer. *m. f.* Micropylar funnel *mp.* Micropyle.

Fig. 8.—Arrangement of filaments at animal pole of egg. *a.* Surface-view. *b.* Longitudinal section.

Fig. 9.—Surface-view of part of base and stem of an outer filament, to show change from polygonal markings to longitudinal striations.

Fig. 10.—Anchor showing longitudinal striations on filament passing up to under surface of anchor, and dark dots on upper surface.

PLATE 12.

Fig. 11.—Transverse section of filament. *col. l.* Columner layer. *hom. l.* Homogeneous layer. *str. l.* Stratified layer.

Fig. 12.—Vertical section of anchor. *h. col.* Heads of columns. *str. l.* Stratified layer.

Fig. 13.—Ventral and two lateral views of embryo.

Fig. 14.—Transverse section of pronephros at last tubule. *b. c.* Body-cavity. *bl.* Blood-vessel. *bo. c.* Bowman's capsule. *gl.* Glomerulus. *s. d.* Segmental duct. tu_1. Last tubule of pronephros.

Fig. 15.—Transverse section of pronephros at second last tubule. tu_2. Second last tubule of pronephros. *tu.* Tubule showing enlargement. Other letters as in fig. 14.